

## REMARKS

In the Office Action, Paper No. 10, received August 26, 2003, the Office Action Summary states that claims 1-60 were pending, claims 1-49 were withdrawn from consideration, and claims 50-60 were rejected. However, it is noted that in the second paragraph on page 2 of the Office Action, the Examiner states "[a]fter reconsideration the restriction between Group I and Group II is withdrawn. Therefore, claims 1-25 and 28 are under consideration in the instant office action." Since claims 1-25 and 28 were not included in the Section 112, second paragraph rejection or the prior art rejections, it is assumed that claims 1-25 and 28 are in condition for allowance. On the other hand, perhaps the Examiner inadvertently overlooked these claims while considering the present application, in which case Applicants respectfully request examination of these claims.

In response to the Office Action, claims 50, 55-56, and 59 have been amended herein and claims 26-27, 29-29, 53, 54, 57, and 60 have been cancelled. In addition, new claims 61-62 have been added to more completely claim the invention. Support for claims 61-62 can be found in the Specification on page 19, lines 13-19, and in Examples 2 and 3. No new matter has been added as a result of these amendments. Reconsideration of the application in light of the above amendments and the following remarks is respectfully requested.

### A. Rejections under 35 U.S.C. § 112, second paragraph, addressed

Claims 50-60 were rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. This rejection is respectfully traversed for the following reasons.

First, the Examiner asserts that the metes and bounds encompassed by "removing a nucleation site" is not clear and the Examiner inquires whether the phospholipid is the nucleation binding site.

Presently pending claim 50 is directed to a method of eliminating or reducing calcification in a biological material by removing a phospholipid calcium nucleation site. As stated on page 6, lines 30-34, the present inventors have found that calcium nucleation correlates with the concentration in a tissue of phospholipid components. Therefore, the invention provides a method of removing phospholipid calcium nucleation sites located in the tissue, so that calcium or other ions are prevented or inhibited from binding to the tissue. Further, as stated on page 9, lines 19-21 of the Specification, the term "nucleation site" is

defined as "that portion of the biological material that forms an association with calcium or other metal ions. The calcium nucleation sites may include phospholipid, protein, and/or polysaccharide components." Accordingly, it is asserted that Specification clearly defines a "phospholipid calcium nucleation site" as a site where a biological material forms an association with calcium and other metal ions, wherein the site comprises phospholipids. Accordingly, it is believed that the meaning of this rejected phrase is clear and definite, and withdrawal of this Section 112, second paragraph rejection is respectfully requested.

Next, the Examiner asserts that while the claims are drawn to a method of "eliminating or reducing the calcification in a biological material," they do not set out sequential method steps needed to achieve the method.

As an initial matter, this rejection has been rendered moot. Without agreeing to the propriety of the rejection, in order to expedite prosecution, claim 50 has been amended herein to recite that the method comprises contacting the biological material with a preparation comprising a surfactant and a denaturing agent. Thus, it is asserted that claim 50 as amended herein and the claims that depend therefrom are clear and definite, and withdrawal of this rejection is respectfully requested. With respect to the rejection of original claim 50, Applicants respectfully note that the discovery that calcium nucleation correlates with the concentration of phospholipid components in a tissue entitles the inventors to broadly claim a method of reducing calcification by removing phospholipid calcium nucleation sites.

**B. Rejections under 35 U.S.C. § 102 (a), (b), or (e) addressed**

Various combinations of claims 50-60 were rejected as being anticipated by Lee et al., Nashef, Vyavahare et al., or Cunanan et al. for the reasons discussed below. As the Examiner is aware, the CAFC has stated that anticipation requires the presence in a single prior art reference of the disclosure of each and every element of the claimed invention, arranged as in the claim. *Lindemann Maschinenfabrik GMBH v. American Hoist & Derrick Co.*, 730 F.2d 1452, 1458 (Fed. Cir. 1984); *Altco Standard Corporation v. Tennessee Valley Authority*, 1 USPQ 1337, 1341 (Fed. Cir. 1986); 774 F.2d 1082 (Fed. Cir. 1985). It is asserted that the above-listed references do not disclose every element of pending claims 50-52, 55-56, 58-59 or every element of newly presented claims 61-62. Accordingly, the Section 102 rejections are respectfully traversed for the reasons discussed below.

1. Lee fails to meet Applicants' claim element of contacting the biological material with a preparation consisting essentially of a surfactant and a denaturing agent.

Claims 50-58 were rejected under 35 U.S.C. § 102(a) or (e) as being anticipated by Lee et al. (U.S. Patent No. 6,008,292). The Examiner first asserts that "any method that removes protein or cellular debris from a biological material would fall within the scope of the claim, by removing the undefined binding sites found in the biological material". The Examiner then asserts that Lee discloses a method of preparing collagenous biological material to inhibit calcification by treating the tissue with Denacol and 20% ethanol as well as treating tissue with a mixture of glutaraldehyde, ethanol and Tween-80 and therefore anticipates the present invention. This rejection is respectfully traversed.

It is asserted that Lee does not expressly or inherently describe all of the elements set forth in claim 50 as amended herein or the claims that depend therefrom. Independent claim 50 as amended now recites a method of eliminating or reducing calcification in a biological material, comprising removing a phospholipid calcium nucleation site contained in the material by contacting the material with a preparation consisting essentially of a surfactant and a denaturing agent. Thus, to anticipate claim 50 and the claims that depend therefrom, Lee must disclose a method of eliminating or reducing phospholipid calcium nucleation sites in a biological material by washing the material with a preparation consisting essentially of a surfactant and a denaturing agent.

In contrast to Applicants' claimed methods, Lee is directed to a method for inhibiting calcification of a fixed collagenous biological tissue by treating the fixed tissue with Denacol (ethylene glycol diglycidyl ether) and ethanol for 120 hours. After measuring the calcium content of the tissue samples following treatment, Lee concludes that ethylene glycol diglycidyl ethers are capable of mitigating tissue calcification (see also Lee's claim 1). It is well known in the art that glycol diglycidyl ethers such as Denacol are crosslinking agents. However, independent claim 50 as amended herein requires that the preparation "consist essentially of" a surfactant and a denaturing agent. It is well known that the transitional phrase "consisting essentially of" limits the scope of the claim to the specified materials or steps and those that do not materially affect the basic and novel characteristic(s) of the claimed invention. Accordingly, the preparation used in the method of claim 50 excludes a crosslinking agent. Therefore, since the Lee requires the use of a crosslinking agent, Lee fails to teach or even suggest all the elements of claim 50 and the claims that depend therefrom.

Accordingly, Lee does not anticipate these claims. Withdrawal of this rejection over Lee is respectfully requested.

2. Nashef fails to meet Applicants' claim element of contacting the biological material with a preparation consisting essentially of a surfactant and a denaturing agent.

Claims 50-58 and 60 were rejected under 35 U.S.C. § 102(b) as being anticipated by Nashef (U.S. Patent No. 4,729,139). The Examiner asserts that Nashef discloses utilizing a disinfecting solution for the processing of bioprosthetic tissue comprising formaldehyde, ethanol and Tween-80, and further asserts that since ethanol and Tween are known to be capable of solubilizing phospholipids and formaldehyde is a well-known sterilization agent, Nashef anticipates the present invention. This rejection is respectfully traversed.

It is asserted that Nashef does not expressly or inherently describe all of the elements set forth in claim 50 or the claims that depend therefrom. As stated above, claim 50 as amended herein recites methods of eliminating or reducing calcification in a biological material, comprising removing a phospholipid calcium nucleation site contained in the material by contacting the material with a preparation consisting essentially of a surfactant and a denaturing agent.

In contrast to Applicants' claimed methods, Nashef discloses a method for incorporating a polymer into a biological tissue to inhibit calcification, which involves fixing the tissue with glutaraldehyde, coupling the tissue with a coupling agent such as a diamine, contacting the coupled tissue with a first monomer solution (e.g., a solution containing acrylic acid) and then exposing the tissue to a polymerization initiator, and then contacting the tissue with a second monomer solution under polymerization conditions.

However, independent claim 50 as amended herein requires that the tissue be contacted with a preparation that consists essentially of a surfactant and a denaturing agent, and therefore the method of the present invention specifically excludes contacting the tissue with a preparation that contains a polymer. Since Nashef fails to teach or even suggest all the elements of claim 50 and the claims that depend therefrom, Nashef does not anticipate these claims. Withdrawal of this rejection over Nashef is respectfully requested.

3. Vyavahare fails to meet Applicants' claim element of contacting the biological material with a preparation consisting essentially of a surfactant and a denaturing agent.

Claims 50-52 and 58 were rejected under 35 U.S.C. § 102(b) as being anticipated by Vyavahare et al. (Circulation, 1997). The Examiner states that Vyavahare discloses the removal of phospholipid from a glutaraldehyde fixed bioprosthetic tissue using an ethanol wash, and thus asserts that Vyavahare anticipates the present invention. This rejection is respectfully traversed.

It is asserted that Vyavahare does not expressly or inherently describe all of the elements set forth in claim 50 or the claims that depend therefrom. As stated above, the claims as amended herein recite a method of eliminating or reducing calcification in a biological material, comprising removing a phospholipid calcium nucleation site contained in the material by contacting the material with a preparation consisting essentially of a surfactant and a denaturing agent.

In contrast to Applicants' invention, Vyavahare discloses a method for preventing calcification of bioprosthetic heart valves using ethanol pretreatments which involves soaking the tissue in ethanol at concentrations greater than 60% for 24 hours to remove membrane lipids and to induce collagen structural changes (page 479, second column, first paragraph; page 480, first column, third paragraph; page 481, second column, second full paragraph). However, Vyavahare does not teach a method of preventing calcification by treating the tissue with a preparation consisting essentially of a surfactant and a denaturing agent. In fact, Vyavahare teaches away from the use of a surfactant, stating on page 487, column 1, second full paragraph that "it has been observed that certain surfactants can result in a severe decrease in the durability of the BPHV". Accordingly, since Vyavahare does not teach every element of claim 50 or the claims that depend therefrom, Vyavahare does not anticipate these claims. Withdrawal of this rejection over Vyavahare is respectfully requested.

4. Cunanan fails to meet Applicants' claim elements of contacting the biological material with a preparation consisting essentially of a surfactant and a denaturing agent.

Claims 50-58 and 60 were rejected under 35 U.S.C. § 102(a) or (e) as being anticipated by Cunanan et al. (U.S. Patent No. 6,214,054) as evidenced by Vyavahare et al. The Examiner states that Cunanan discloses a method of preparing bioprosthetic tissue using a combination of formaldehyde, ethanol and Tween for the processing of the tissue, and Vyavahare provides evidence that ethanol treatment is effective at removing phospholipids from tissue. From this the Examiner concludes that Cunanan anticipates the present invention. This rejection is respectfully traversed.

It is asserted that Cunanan does not expressly or inherently describe all of the elements set forth in claims 50-58 and 60. As stated above, claim 50 amended herein recites a method of eliminating or reducing calcification in a biological material, comprising removing a phospholipid calcium nucleation site contained in the material by contacting the material with a preparation consisting essentially of a surfactant and a denaturing agent.

In contrast to Applicants' invention, Cunanan discloses a method for preparing a bioprosthesis in a manner that mitigates post-implantation calcification of the bioprosthesis. The method involves immersing the tissue in a denaturant/surfactant/crosslinking agent mixture for 2 to 24 hours and then rinsing the tissue. However, independent claim 50 as amended herein requires that the preparation "consist essentially of" a surfactant and a denaturing agent, and therefore specifically excludes contacting the tissue with a preparation that contains a crosslinking agent. Accordingly, since Cunanan does not teach every element of 50-58 and 60, Cunanan does not anticipate these claims. Withdrawal of this rejection over Cunanan is respectfully requested.

**C. Rejections under 35 U.S.C. § 103(a) addressed**

Claims 50-60 are rejected under 35 U.S.C. § 103(a) as being unpatentable over Nashef in view of Mirsch et al. (U.S. Patent No. 6,121,041). The Examiner states that Nashef teaches utilizing a disinfecting solution for the processing of bioprosthetic tissue comprising formaldehyde, ethanol and Tween-80, and further states that the process taught by Nashef achieves reduced calcification in bioprosthetic tissue. The Examiner then states that Nashef fails to teach the addition of a phospholipase, and cites Mirsch for teaching that lysed cells can provide a source of enzymes and other material effective for decellularization. From this the Examiner concludes that it would have been obvious to one of ordinary skill in the art to utilize the enzymes produced by a microorganism and combine them with the cleansing process taught by Nashef. This rejection is respectfully traversed.

As a first matter, claim 50 as amended herein requires that the tissue be contacted with a preparation that consists essentially of a surfactant and a denaturing agent, and therefore the method of the present invention specifically excludes contacting the tissue with a preparation that contains a phospholipid. Accordingly, the use of Mirsch in the rejection of claims 50-52, 55-56, and 58 is moot, since the transitional phrase "consisting essentially of" excludes a phospholipase from the preparation that contacts the tissue. For this reason alone, the rejection of claims 50-52, 55-56 and 58 falls.

With respect to claim 59, it is asserted that the combination of Nashef and Mirsch does not render claim 59 obvious. As stated above, Nashef discloses a method for incorporating a polymer into a biological tissue to inhibit calcification, which involves fixing the tissue with glutaraldehyde, coupling the tissue with a coupling agent such as a diamine, contacting the coupled tissue with a first monomer solution (e.g., a solution containing acrylic acid) and then exposing the tissue to a polymerization initiator, and then contacting the tissue with a second monomer solution under polymerization conditions. However, claim 59 as amended herein requires that the tissue be contacted with a preparation that consists essentially of a surfactant, a denaturing agent, and a phospholipase, and therefore specifically excludes contacting the tissue with a preparation that contains a polymer.

Mirsch discloses a method of inoculating a tissue with a solution containing microorganisms, wherein the microorganisms are selected to produce compounds that process the tissue. Mirsch states that the tissue can subsequently be treated to remove or inactivate the microorganisms by "ionizing radiation, ultraviolet irradiation, antibiotics and chemical exposure" (see Mirsch claim 1). However, Mirsch is silent with respect to a method of removing calcium nucleation sites for the microorganisms, and further Mirsch does not specifically disclose phospholipases. In any event, even if Mirsch did disclose a phospholipase, the combination of Nashef and Mirsch would not render claim 59 obvious, since as stated above claim 59 excludes the addition of a polymer in the solution used to contact the tissue. Withdrawal of this rejection is respectfully requested.

**D. Obviousness-type double patenting rejection addressed**

Claims 50-58 and 60 were rejected under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 1-27 of U.S. Patent No. 6,214,054 in view of Vyavahare et al. The Examiner asserts that although the conflicting claims are not identical, they are not patentably distinct from each other because the method disclosed in U.S. Patent No. 6,214,054 will result in the removal of phospholipid from the fixed bioprosthetic tissue, as evidenced by Vyavahare et al. where phospholipid removal was achieved with ethanol treatment of glutaraldehyde fixed bioprosthetic tissue.

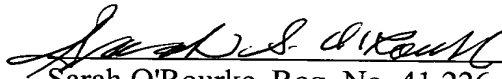
A Terminal Disclaimer with respect to the 6,214,054 patent will be submitted when patentable subject matter is determined.

### CONCLUSION

Accordingly, in view of the above amendments and remarks, it is submitted that this application is now ready for allowance. Early notice to this effect is solicited. If in the opinion of the Examiner, a telephone conference would expedite the prosecution of the subject application, the Examiner is invited to call the undersigned at the telephone number listed below. The fee associated with the filing of a three month time extension accompanies this response. No additional fee is believed due as a result of filing this Amendment. However, should any additional fees be due the Examiner is authorized to charge Deposit Account No. 50-1123.

Respectfully submitted,

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Dated

  
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